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Effects of Propranolol on Renin Release during Chronic Thoracic Caval Constriction or Acute Renal Artery Stenosis in Dogs (39366)

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The mechanisms which regulate renin release in dogs with chronic thoracic caval constriction (1) or with suprarenal aortic constriction (2-4) have been studied under acute conditions during anesthesia. In dogs with chronic thoracic caval constriction the renal nerves play an important modulatory role in renin release although they are not essential since as much as an eightfold increase in PRA (1) was present after renal denervation. An increase in renin secretion occurred following acute suprarenal aortic constriction in adrenalectomized, anesthetized dogs with a denervated, nonfiltering kidney (4). This latter study eliminated the influence of increased circulating epinephrine but not norepinephrine, and the affects of neither catecholamine were excluded in the studies of the dogs with chronic caval constriction. The present observations with propranolol were made to determine the effect of blockade of the β-receptor mechanism for renin relesae in these two experimental situations.

Materials and methods. Experiment 1. Chronic response to d,1-propranolol in dogs with thoracic inferior vena cava constriction. Six female mongrel dogs weighing from 19-24 kg were placed in metabolic cages and urine was collected daily for determination of renal sodium and potassium excretion. The animals were maintained on a diet containing 60 and 45 mEq/day of sodium and potassium, respectively. Arterial blood pressure and heart rate were measured by percutaneous puncture of the femoral artery with a 22-gauge needle attached to a Statham pressure transducer and recorded on a Hewlett-Packard 7702B recorder. All dogs were started on oral d,1-propranolol (240 mg twice/day) at 8:00 AM and 4:00 PM at least two days before the thoracic inferior vena cava was constricted under sterile conditions to produce experimental ascites (5). Six to 8 days after caval constriction, d,1-propranolol administration was stopped and the dogs allowed to recover. Unless otherwise stated, all blood samples were taken from the jugular vein in the morning 16 hr after 4:00 PM dose of the previous day of d,1-propranolol.

Experiment 2: Acute response to d,1-propranolol in conscious dogs with caval constriction. Six to 8 days after caval constriction, four of the dogs of experiment 1 were brought to the laboratory and their bladders were catheterized and washed with distilled water. A control urine sample (1 hr) and a blood sample for PRA and plasma propranolol determination were taken before the morning dose of d,1-propranolol was administered. Renal sodium excretion, PRA and plasma propranolol levels were measured every hour for 5 hr following propranolol administration. During the experiment the dogs lay quietly on the floor.

Experiment 3: The effects of d,1-propranolol in conscious dogs with acute renal artery constriction. An inflatable cuff was placed around the left renal artery of four female mongrel dogs (19-21 kg) under sterile conditions; a tube extended to the outside of the chest wall. The dogs were allowed to recover for at least 7 days before the acute experiment; they were maintained on a standard diet as described above. For the acute experiment, arterial pressure was measured through an indwelling femoral artery catheter. A saphenous vein was cannulated for infusion of d,1-propranolol. Blood samples for PRA, plasma electrolytes, and plasma propranolol concentration were obtained from either a jugular or a saphenous vein. After the vessels were cannulated, a saline infusion (0.5 ml/min) was begun at least 30 min before the first blood samples were taken. After two control samples (15 min apart), the renal artery cuff was inflated with 0.5-1.5 ml of water to constrict the vessel. Blood samples were taken 30 and 45

min after constriction. The constriction was then released and the dogs allowed to recover before samples were taken after 60 and 75 min. d,l-Propranolol was then infused at 0.2 mg/kg/hr and a propranolol blood sample was taken after 30 min. The renal artery cuff was inflated to exactly the same degree by use of the same amount of water and samples taken 30, 90, 150, 210, and 270 min during propranolol infusion and renal artery stenosis. Throughout the experiment the animals lay quietly on the floor.

Analytical methods. Venous blood samples for the determination of PRA were placed in chilled test tubes containing 0.1 ml of 10% ethylenediaminetetraacetic acid for each 10 ml of blood. The samples were centrifuged and plasma was frozen until it was prepared for assay by the method of Schneider *et al.* (6). PRA was determined by measuring the pressor response in the pentobarbital-anesthetized, pentolinium-blocked rat in comparison with known amounts of angiotensin II (Hypertensin, Ciba). PRA is expressed as nanograms angiotensin II/ml generated during 3 hr of incubation. Venous blood samples for the determination of plasma electrolytes and propranolol concentrations were obtained and placed in chilled test tubes containing 0.05 ml of heparin per 10 ml of blood and held in an ice bath until centrifuged. The plasmas were decanted and refrigerated until analyzed. Plasma and urine electrolytes were determined by flame photometry. Plasma propranolol levels were assayed by Ayerst Laboratories (Montreal, Canada) by the fluorometric method of Shand *et al.* (7).

Statistical analysis of paired data was performed by Student's *t* test.

Results. Experiment 1: Chronic response to d,l-propranolol in dogs with thoracic inferior vena cava constriction. The response of six dogs to caval constriction during chronic propranolol administration is presented in Fig. 1. In all six animals high circulating levels of plasma propranolol were obtained 3 to 8 hr after the morning dose of the drug. However, 16 hr after the evening dose, plasma levels were low. Heart rate was reduced significantly with propranolol administration from 133 ± 7 (SEM) beats/min to 95 ± 6 beats/min ($P < 0.005$). Heart rate remained low during caval constriction and propranolol treatment (83 ± 8 beats/min) ($P < 0.025$) and during recovery increased to 131 ± 10 beats/min ($P < 0.0125$). These values for heart rate were recorded in the morning 16 hr after the evening dose of propranolol. PRA increased significantly after constriction of the inferior vena cava although d,l-propranolol was administered ($P < 0.025$). During the last two days of caval constriction and propranolol administration, PRA fell toward the control level ($P < 0.05$) but did not reach this level ($P < 0.025$). After d,l-propranolol administration was stopped PRA rose to 20 ± 3 ng angiotensin II/ml ($P < 0.05$). Arterial pressure fell after caval constriction and d,l-propranolol administration from 143 ± 7 to 128 ± 7 mm Hg ($P < 0.01$) and increased during recovery to 145 ± 6 mm Hg ($P < 0.025$). In all animals renal sodium excretion decreased during the second day of thoracic caval constriction and remained low throughout the remainder of the experiment. Ascites formation continued unabated throughout d,l-propranolol administration and caval constriction.

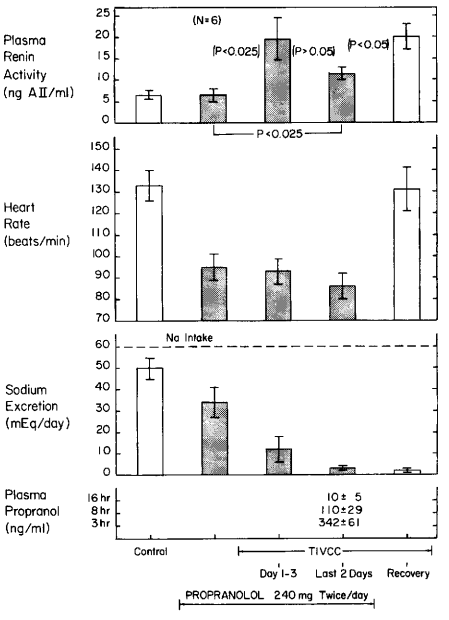

FIG. 1 Chronic response to d,l-propranolol in six dogs with chronic thoracic caval constriction. Abbreviations are same as in Fig. 2.

There was no change in plasma potassium concentration during the administration of d,1-propranolol.

Experiment 2: Acute response to d,1-propranolol in dogs with thoracic caval constriction. Since propranolol reached such low levels 16 hr following the afternoon dose, it was decided to observe the changes in PRA and renal sodium excretion during high levels of circulating propranolol. The experiment was designed to determine if PRA returned to normal and if sodium excretion increased transiently when the circulating level of propranolol was high. During the control observations (18–21 hr after the afternoon dose of d,1-propranolol), the plasma level of propranolol was undetectable in two of the four dogs. During this control period, PRA was 10 ± 2 ng angiotensin II/ml. in comparison with the 80-ng level found in dogs with caval constriction but not receiving d,1-propranolol (1), and renal sodium excretion was low (2–3 μEq/min). PRA and renal sodium excretion were unchanged for 5 hr after a 240-mg oral dose of d,1-propranolol; at 3 and 5 hr the plasma propranolol levels were 210 and 130 ng/ml, respectively.

Experiment 3: The effects of d,1-propranolol on acute renal artery constriction in conscious dogs. The response of four dogs to renal artery stenosis before and during an intravenous infusion of d,1-propranolol is presented in Fig. 2. Before propranolol administration, arterial pressure increased during constriction from 120 ± 3 to 136 ± 6 mm Hg ($P < 0.05$) and decreased during recovery after release of the constriction to 119 ± 4 mm Hg ($P < 0.025$). PRA increased with renal arterial constriction from 6 ± 1 to 12 ± 4 ng angiotensin II/ml ($P < 0.05$) and fell during recovery (6 ± 1 ng angiotensin II/min) ($P < 0.05$). Heart rate fell from 110 ± 10 to 84 ± 10 beats/min ($P < 0.025$) during the first 30 min of d,1-propranolol infusion before reconstriction of the renal artery and remained low during d,1-propranolol infusion. Upon reconstriction of the renal artery during d,1-propranolol, arterial pressure increased from 119 ± 4 to 146 ± 9 mm Hg ($P < 0.025$). Again, PRA more than doubled during renal artery stenosis ($P < 0.005$). Neither the increased arterial pressure nor PRA was attenuated by d,1-propranolol infusion during renal artery constriction. However, during d,1-propranolol infusion before renal artery stenosis PRA decreased significantly from the last recovery period from 5.9 to 4.0 ± 1 angiotensin II/ml ($P < 0.05$). Plasma potassium fell significantly from 4.6 ± 0.1 to 4.1 ± 0.1 mEq/liter ($P < 0.05$) after 5 hr of propranolol infusion, but was unchanged during the first 4 hr.

Discussion. Constriction of the thoracic inferior vena cava in dogs is a potent stimulus for the hypersecretion of renin, hyperaldosteronism, salt, and water retention, and ascites formation (1, 5, 8, 9). Studies during chronic renal denervation (1) have revealed a 50% drop in PRA from a 16-fold to an 8-fold elevation above normal without any detectable change in renal sodium excretion or ascites formation. Also, Witty *et al.* (1) observed an acute decrease in renin secretion during intravenous infusion of propranolol in anesthetized dogs with caval constriction but not to the control level. In the

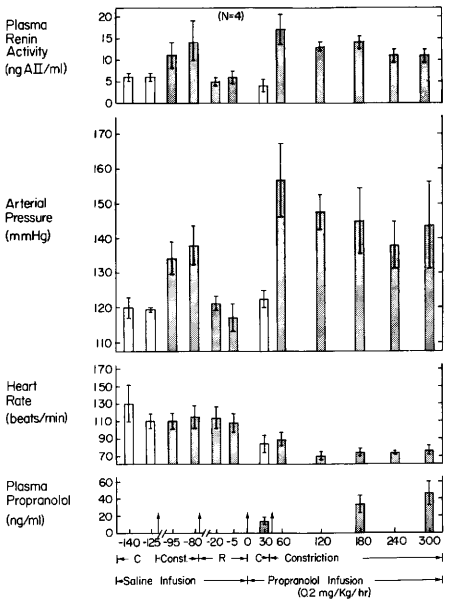

FIG. 2. Acute response to renal artery constriction before and during d,1-propranolol administration in dogs. Abbreviations of C, Const., and R are, respectively, for control, constriction, and recovery. The open bars are for values determined during the control periods.

present study, chronic daily propranolol administration failed to block the increase in PRA in conscious dogs in response to thoracic caval constriction; PRA increased from an average control value of 6 ± 1 ng of angiotensin/ml before caval constriction to a maximal level of 23 ± 3 ng angiotensin/ml ($P < 0.025$) after 4–5 days of propranolol administration. These are the first such data in conscious dogs with caval constriction in which actual measurements of plasma propranolol have been made and there is evidence of complete β-blockade. However, this level of PRA was significantly lower ($P < 0.001$) than the high PRA of 80 ng of angiotensin/ml observed previously (1) in conscious intact dogs with caval constriction studied under similar conditions. Nevertheless, during propranolol PRA was high enough to produce almost complete retention of sodium and ascites formation. Plasma propranolol levels during the first 8 hr after the morning dose were as high or higher than those observed to produce complete β-adrenergic blockade (10–12). The low heart rate present before the 8:00 AM dose of propranolol also indicates that β-blockade was still present 16 hr after the last dose given at 4:00 PM on the previous afternoon. In four dogs studied acutely after 6–8 days of propranolol administration, the average PRA was 10 ± 2 ng angiotensin/ml before the 8:00 AM dose of propranolol. These findings suggest that β-blockade was essentially complete at 16 hr after the last 4:00 PM dose of propranolol from the previous day. Also, renal sodium excretion during the 5 hr remained at the low rate characteristic of dogs with thoracic caval constriction. It should be emphasized that there is no evidence that d,1-propranolol exerts a local anesthetic action to block renin release because the d and 1 isomers of propranolol are equally potent in stabilizing the cell membrane (13) but only 1-propranolol blocks renin release (14). These observations show that nonadrenergic mechanisms are involved in the hypersecretion of renin in dogs with chronic caval constriction.

The role of the adrenergic nervous system in mediating the increase in renin secretion following renal artery stenosis was also studied. The important feature of the experiment is that there was no attenuation by propranolol of the increases in PRA and arterial pressure secondary to renal artery constriction. Plasma propranolol levels increased progressively during the 5-hr infusion to reach an average level of 46 ± 14 ng/ml; this level has been reported previously (10–12) to achieve complete β-blockade. These data, therefore, give no indication that a β-adrenergic receptor mediates the increase in renin release following renal artery stenosis. The findings agree with an earlier report of the effects of renal artery constriction in anesthetized dogs by Winer (15) and a more recent preliminary study in two conscious dogs following renal artery stenosis (16). It seems likely that the increase in renin secretion following renal artery constriction is mediated at least in part by the renal vascular receptor (2, 3). Evidence for involvement of this receptor was provided by Blaine, *et al.* (2, 3) who showed that acute aortic constriction increased renin release in adrenalectomized dogs with a denervated, nonfiltering kidney.

Summary and conclusions. Dogs were given d,1-propranolol before and after thoracic inferior vena cava constriction (TIVCC) or renal artery constriction (RAC) to determine if the increase in plasma renin activity (PRA) and associated biological activity could be suppressed. Oral propranolol administration (240 mg twice daily) was begun at least 2 days prior to TIVCC in six dogs; heart rate was reduced from 133 to 95 beats/min but PRA did not change with propranolol administration. After TIVCC and during continued propranolol administration, daily renal sodium excretion fell from an average value of 50 to less than 3 mEq sodium/day and PRA was elevated two- to fourfold. With prolonged propranolol adminstration during TIVCC, PRA returned toward normal levels but renal sodium excretion remained low. At this time, hourly measurements of sodium excretion showed no change after a 240-mg oral dose of propranolol although very high plasma levels of propranolol were achieved; also, PRA was unchanged and low. The effects of RAC were studied before and during an intravenous propranolol infusion (0.2 mg/kg/hour) in conscious animals. Before propranolol

administration, arterial pressure increased during RAC from 120 to 136 mm Hg and PRA doubled. Propranolol infusion lowered heart rate from 110 to 84 beats/min, but arterial pressure and PRA were not attentuated by propranolol during RAC. The data indicate that non-beta-adrenergic mechanisms are involved in renin release during TIVCC and RAC.

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